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# KARAKTERIZACIJA PATOTIPOVA GLJIVICE *Passalora fulva* (Cooke) U.Braun&Crous UZROČNIKA BARŠUNASTE PLIJESNI LISTA RAJČICE U HRVATSKOJ

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*Disertacija* <sup>(2)</sup>

Rajčica je jedna od povrtnih kultura čija se proizvodnja povećava iz godine u godinu, i u svijetu i u Hrvatskoj. U ovom su radu prikazana istraživanja i njihovi rezultati o uzročniku bolesti baršunaste plijesni lista rajčice - *Passalora fulva* (Cooke) U.Braun & Crous (sin. *Cladosporium fulvum* (Cooke), koja utječe na prinos i kakvoću rajčice. Osim gospodarske štete, baršunasta plijesan zbog intenzivne sporulacije može pogodovati razvoju alergijskoga bronhitisa pa time, u višekratnoj berbi, narušiti zdravstveno stanje proizvođača rajčice. Tijekom istraživanja korišteni su sljedeći materijali i metode rada: prikupljanje uzoraka biljnoga materijala zaraženoga baršunastom plijesni lista rajčice i ocjena jačine zaraze, izolacija i determinacija patogena, određivanje patotipova biotestom, ispitivanje osjetljivosti različitih genotipova rajčice na zarazu *P. fulva*, molekularna identifikacija i utjecaj eteričnih ulja na klijavost konidija *in vitro*. Vizualni pregledi nasada rajčice u zaštićenome prostoru i na otvorenome provedeni su u razdoblju 2006.- 2011. Prikupljeno je 17 uzoraka u zaštićenome prostoru na 14 lokaliteta u 10 županija (Istarska, Koprivničko-križevačka, Krapinsko-zagorska, Međimurska, Osječko-baranjska, Primorsko-goranska, Varaždinska, Virovitičko-podravsko, Zadarska i Zagrebačka). Znakovi zaraze zabilježeni su na šest genotipa rajčice: Belle, Jeremy, Minaret, Amaneta, Gravitet i pokusni genotip. Na otvorenome *P. fulva* nije utvrđena. Za izolaciju i opis *P. fulva* korištene su četiri različite podloge: PDA (krumpir – dekstrozni agar), rajčica – agar, mrkva – agar i V8 - agar. Mjerenjem konidija utvrđeno je da je veličina konidija na mrkva–agar podlozi najbliža veličini konidija izmjerenih sa zaraženog lista rajčice. Biotestom je po prvi put u Hrvatskoj utvrđeno prisustvo patotipova *P. fulva*, Cf – 0 i Cf – 5. Metodom lančane reakcije polimeraze (PCR) potvrđen je patotip Cf–2. Ostali patotipovi dobiveni biotestom nisu potvrđeni sekvenciranjem, jer dobivene sekvence nisu bile zadovoljavajuće za interpretaciju rezultata. Molekularnom metodom potvrđena je gljiva *P. fulva* u svim testiranim uzorcima. Prvi je put u Hrvatskoj obavljeno ispitivanje osjetljivosti genotipova rajčice na zarazu *P. fulva*. Osjetljivi genotipovi su Saint Pierre, Volovsko

srce, Marglobe i S. Marzano. Prvi je put u Hrvatskoj, prema dostupnoj literaturi i u svijetu, ispitivan utjecaj 6 eteričnih ulja na klijanje konidija *P. fulva in vitro*. Najučinkovitije je bilo ulje klinčićevca pri koncentraciji 10 µl/ml, kod kojeg se postotak iskljajalih konidija kretao između 9 i 21%.

Ključne riječi: rajčica, baršunasta plijesan lista rajčice, *Passalora fulva*, hranjive podloge, patotipovi, PCR, osjetljivost genotipova rajčice, eterična ulja, Hrvatska

## THE CHARACTERISATION OF FUNGI *Passalora fulva* (Cooke) U.Braun&Crous PATHOTYPES, CAUSER OF TOMATO LEAF MOLD IN CROATIA

*Doctoral thesis*

Tomato is considered one of the most widely grown vegetable crops in the world and production of it is rising every year in Croatia as well as in the world. This research paper shows results on tomato leaf mould caused by *Passalora fulva* (Cooke) U.Braun & Crous (syn. *Cladosporium fulvum* (Cooke)), which can cause significant reduction of yield and quality of tomato. Other than economic loss, tomato leaf mould can be the cause of allergies because it produces lots of spores. During research, the following materials and methods have been used: collecting samples of plant material infected with tomato leaf mould and assessments of disease intensity, isolation and determination of the pathogen, detecting pathotypes using biassay, susceptibility testing of different tomato genotypes infected with *P. fulva*, molecular identification and testing the effects of essential oils *in vitro* on conidia germination. Visual controls of greenhouse and outdoor growing tomato were conducted during the period from 2006 to 2011. Samples (17) from 14 localities in 10 counties (Istra County, Koprivnica-Križevci County, Krapina-Zagorje County, Međimurje County, Osijek-Baranja

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County, Primorje-Gorski Kotar County, Varaždin County, Virovitica-Podravina County, Zadar County and Zagreb County) were collected. Symptoms were detected on 6 different tomato genotypes: Belle, Jeremy, Minaret, Amaneta, Gravitet and test genotype. Outdoors, *P. fulva* was not found. For isolation and determination of *P. fulva* 4 different growing media were used: PDA (potato – dextrose agar), tomato – agar, carrot – agar and V8 – agar. By measuring the size of conidia on carrot – agar it was determined that they were the closest in size to the conidia from infected tomato leaf. Bioassay for the first time in Croatia confirmed presence of two different physiological pathotypes of *P. fulva*, Cf – 0 and Cf – 5. Other pathotypes obtained by bioassay were not confirmed by sequencing because the sequences obtained were not satisfactory for the results interpretation. Primary PCR confirmed the fungus *P. fulva* in all samples tested. Susceptibility testing of tomato genotypes on *P. fulva* infection was done for the first time in Croatia. Susceptible genotypes were Saint Pierre, Volovsko srce, Marglobe and S. Marzano. For the first time in Croatia, and according to the literature available probably in the world as well, the effects of essential oil on conidial germination of *P. fulva in vitro* were tested. The most effective oil was clove oil in the concentration of 10  $\mu$ l/ml where the percentage of germinated conidia ranged between 9 and 21%.

Key-words: tomato, tomato leaf mould, *Passalora fulva*, growing media, pathotypes, PCR, susceptibility testing of tomato genotypes, effects of essential oils, Croatia